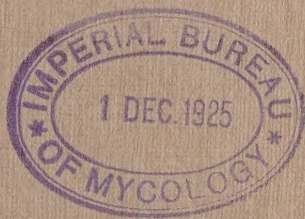


Basisporium Dry Rot of Corn

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IOWA STATE COLLEGE OF AGRICULTURE
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BOTANY AND PLANT PATHOLOGY
SECTION



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SUMMARY

Basisporium gallarum was first found on corn from Bulgaria in 1911 by Bubak and by Arzberger in Ohio in 1913. *Coniosporium gecevi* Bubak is identical with *Basisporium gallarum* Moll. The common name of *Basisporium* dry rot is suggested for the disease caused by this organism.

The dry rot of corn, caused by this fungus, was very prevalent in Iowa in 1923, causing an average damage to the crop of 9.1 percent, while in a few fields from 50 to 60 percent of the ears were infected. The damage consists of moldy and light ears, reduced stand and weak plants the succeeding year.

Heavy precipitation in August and September, when the crop is maturing, favors the development of the disease. Studies to date indicate that *Basisporium* dry rot is markedly dependent upon excessive moisture conditions when the crop is maturing, for its destructive development.

Basisporium gallarum attacks the shanks, husks and stalks of corn. The shanks particularly are weakened and break easily. On the ears the fungus is visible at the butt and base of the kernels. The black spores of the fungus can be readily seen with the unaided eye. The kernels are affected in varying degree, some only slightly, while in others the embryo is killed.

The spores germinate poorly or not at all in water. They germinate readily in tomato or orange juice. Such juice neutralized to litmus failed to produce germination. Plant tissues in the same atmosphere with the drop cultures also stimulate germination.

This action is due to the CO_2 given off by the tissues. Carbon dioxide in small amounts produces a like effect.

The maximum, optimum and minimum temperatures for spore germination are 35°C ., 25°C ., and 15°C ., respectively. The optimum temperature for mycelial growth is 25°C ., the good growth takes place between 20°C . and 35°C .— 40°C . and 10°C . inhibit growth. Sporulation may take place between 20°C . and 35°C . The fungus readily winters over in its conidial stage.

Basisporium gallarum grows well on a wide range of media, but prefers especially media rich in nitrogenous material. On the corn kernel, the fungus destroys the embryo before it does the starchy endosperm.

Corn is most susceptible to attack during time of germination of the seed and late stages of maturity.

Inoculation of growing roots and stalks yielded negative results. *Basisporium* dry rot does not spread readily in cribbed corn.

In 1923 early varieties became generally infected while late varieties remained free.

BASISPORIUM DRY ROT OF CORN

By L. W. DURRELL*

During the investigations of corn diseases in progress at this station, the unusual prevalence of a dry rot due to a rather inconspicuous fungus, *Basisporium gallarum*, was observed in 1923. Nine percent of the ears in seven experimental fields in different sections of the state were partially or wholly destroyed by *Basisporium*. In many fields in sections of the state where the rainfall was heaviest during August and September, from 50 to 60 percent of the ears were infected. In addition to reducing the yield and quality of the crop in 1923, *Basisporium* also attacked much of the seed, causing marked reduction in stand.

The economic significance of this dry rot and the lack of knowledge about it, coupled with the repeated request for information from corn growers, suggested a study of this disease. To date, the causal organism has been identified, the symptoms and effect of the disease followed, and the relation of environmental conditions and maturity of the host to its seasonal prevalence investigated. In addition, spore germination studies leading to the determination of carbon dioxide as a stimulating agent, its relation to mode of infection and effect on seed are reported.**

PREVIOUS RECORD OF *BASISPORIUM GALLARUM*. MOLL.

Basisporium gallarum was first described by Molliard (11) in 1902 as occurring on dead larvae of *Lipara lucens* in galls made by this insect in *Phragmites communis*. Later, Dale (6) isolated an organism from soil in 1913 which she referred to Molliard's description and discussed its relationship to the closely allied genera, *Rhizocladium* and *Trichosporium*.

Meanwhile, Bubak (4) in 1911 had described a fungus from corn plants sent him from Bulgaria and named it *Coniosporium gecevi* Bubak.

During the same year, Arzberger (1) began work in Ohio on a disease of corn which he termed "cob rot" caused by *C. gecevi*. He published his data in 1913. The disease described by Arzberger is the same as the one reported in this paper, according to comparisons made with specimens of the rotted cob fungus, which were supplied by Mr. Arzberger. He had sent specimens of his fungus to Bubak, who identified it as *Coniosporium gecevi*

*The writer acknowledges his indebtedness to Dr. I. E. Melhus for advice and assistance during the work and in the preparation of the manuscript; also to Dr. J. C. Gilman for assistance in determination of the causal organism.

**This bulletin is issued as a progress report because Dr. Durrell has left the Iowa Station. The investigations are being continued.

Bubak. The fungus was cultured artificially, but Arzberger was unable to obtain infection on seedlings or older growing plants. He found, however, that it readily attacked sterilized green ears or moistened crib corn. He also found it on a single head of barley which had previously been killed, but did not find it on living plants.

Ramsey (13) recently isolated the fungus from rotted tomatoes and determined its pathogenicity on that fruit in storage. He refers his organism to *Basisporium gallarum* Moll. He reports in a foot-note that Jenkins has found this fungus on cultures from corn, wheat and dewberries.

From a comparison of specimens, descriptions and published figures, it is evident that *Coniosporium gecevi* Bubak and *Basisporium gallarum* Moll. are identical. The presence of the characteristic basal cell in the conidiophore as opposed to the simple conidiophore found in the genus *Coniosporium*, places it logi-

cally in the genus *Basisporium*. It should also be noted that *Basisporium gallarum* Moll. antedates *Coniosporium gecevi* Bubak by nine years.

COMMON NAME.

It has already been mentioned that Arzberger called the disease in question "cob rot" caused by *Coniosporium gecevi*, a name that in some ways is quite descriptive, but not always readily distinguished from other rots on the ear and cob, i.e., dry rots caused by *Diplodia zae* and *Gibberella saubinetii*. Such being the case, the name *Basisporium* dry rot, will be used in this publication.

SYMPTOMS OF BASISPORIUM DRY ROT

Altho Bubak and Arzberger both studied *Basisporium* dry rot of corn, their descriptions of its



Fig. 1. Corn shank showing surface peppered with spores of *Basisporium*.

symptoms are not fully adequate. There are four places where it is characteristic in appearance. They are: on the shanks, husks and stalks so that the disease may be known in the field at harvest-time; on the ears so that it may be recognized in the crib or in selecting seed ears; on the kernels as confirming it on the ear; and on the germinator so that ears having the disease may be discarded. The symptoms will be discussed in the order named.

Shanks, Husks and

Stalks.

Basisporium dry rot on the shanks, husks and stalks is indicated by the presence of the black spores of the fungus on the surface of the affected part. There is little discoloration, usually only a little darkening of the epidermis which is covered with the fruiting bodies of the fungus. This appearance is always within the leaf sheath. On the unexposed portions of the ear-shanks where conditions of extreme moisture prevail, the surface shows a dusty, black appearance (fig. 1). The spores, being black, round and clustered, give

the surface on which they are borne a black-powdery, or grayish-black appearance. The disease has never been observed on leaves or roots, but it is occasionally found on the nodes, particularly the shank nodes (fig. 2). Accompanying the external symptoms of the fruiting bodies of the fungus, the shank of the ear is usually retted so that the vascular bundles appear as strands of rope (fig. 2). This retting so weakens the shank that the ears readily snap off at harvest and are often knocked to the ground instead of dropping into the machine where harvesting machines are used.

On the ears. While *Basisporium* dry rot is readily recognized on the stalks and shanks, it is probably most characteristic and



Fig. 2. Shank of ear retted by action of *Basisporium*. Fungus fruiting over surface of nodes.

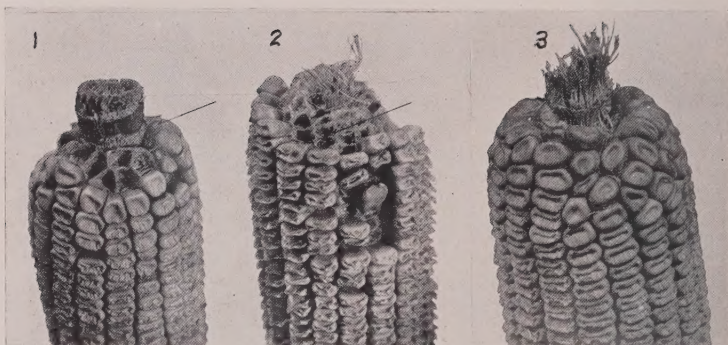


Fig. 3. Ears one and two showing butts attacked by *Basisporium*. Fungus fruiting causes black appearance of chaff and cob at attachment of kernels. Ear three shows retted shank caused by action of *Basisporium*.

evident on the ears. Here the fungus grows on the cob and over the base of the kernels (fig. 3, ears No. 1 and 2), fruiting profusely and thereby giving these parts the black or grayish appearance noted above in the case of the shanks. The mycelium is sparse and inconspicuous. It occurs very abundantly, however, in the cob tissues and often in the furrows between the rows of kernels. The butt of the cob also shows the characteristic retting already described on the shank. When the infection is quite general, the kernels are very loose on the cob and the ear is easily broken.

On the kernels. The symptoms of *Basisporium gallarum* on the kernels vary markedly. It may be seen fruiting only on the tip cap, and thereby discoloring it or the fungus may extend up the kernel and cover its surface with the black spores (fig. 4).

Microscopic examination of the surface of such an infected kernel reveals a fine web of white mycelium with many black spherical spores sprinkled over it (fig. 5). The effect on the kernel is variable. In some instances only the dead tissue of the tip cap is affected and microscopic examination shows no mycelial invasion beyond the suberin layer which protects the embryo. In other cases, the embryo itself is invaded and markedly discolored (fig. 6).

Kernels infected with this rot, unlike those attacked by *Diplodia zeae* or *Gibberella saubinetii*, are not covered with mycelial growth when they germinate on the germinator. Only a faint web of white mycelium is obvious. Kernels only slightly attacked germinate, while others with deeper infection are very weak or dead.

DAMAGE TO CORN CAUSED BY BASISPORIUM DRY ROT

Basisporium dry rot was prevalent and destructive in the fall of 1923. In the fall of 1924, however, it was unusually prevalent. The average in experimental fields in seven counties in representative sections of the state was 9.1 percent of the ears infected. A considerable portion of the infected ears were left in the field, while others were gathered with the sound ones. In the latter instances, the moldy ears influenced the quality and grade on the market. Many fields in the wet areas of the state had a greater amount of infection, some having as much as 50 to 60 percent of the ears attacked.

From the symptoms previously described, the damage to corn caused by *Basisporium gallarum* is obviously two-fold: first, rotting of the ear and shank resulting in moldy and light weight ears; secondly, a dry rot of the kernels which injures the germination of seed corn.

Fig. 4. Badly diseased kernel showing the mycelium and profuse sporulation of *Basisporium* on surface.

In table I is given the weight of 199 healthy ears and 112 slightly attacked by *Basisporium* dry rot. The ears were grouped into four classes on the basis of length as shown in table I. The healthy ears averaged from 14 to 47 grams heavier than the infected ones.

In another series of 169 ears the uninfected ears weighed on an average 282 grams dry weight, and the visibly infected 219 grams dry weight, a difference of 22 percent. This decrease in weight was partly the result of the action of the fungus on the cob and kernels, and in part was due to the poor de-

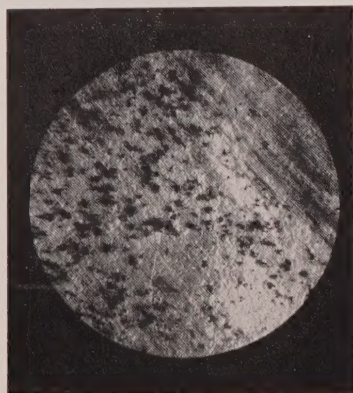


Fig. 5. Micro photograph of surface of diseased corn kernel showing groups of black spores and fine white mycelium of *Basisporium*.

TABLE I. DRY WEIGHT OF BASISPORIUM DRY ROT AND HEALTHY EARS OF CORN.

Infected ears			Healthy ears		
Number of ears weighed	Length inches	Average weight grams	Number of ears weighed	Length inches	Average weight grams
112	7	200	199	7	239
...	8	221	...	8	268
...	9	270	...	9	302
...	10	335	...	10	349

velopment of the ear which followed the shank injury by the fungus during maturity. Markedly infected ears are quite worthless for feed and react unfavorably on the grade when marketed.

From the standpoint of seed corn, the presence of the fungus on a kernel is not necessarily an indication that the kernel is dead. Kernels quite black with spores may germinate, while plantings of the embryos of such kernels have been found free of the organism. The fungus on slightly diseased kernels penetrates the tip cap of the kernel pervading all its tissue. On those kernels where penetration is deeper than this, the embryo is consumed rather than the starchy endosperm, (fig. 6).

In order to determine the degree of this injury, kernels from 32 diseased ears were planted in the greenhouse and the stand and condition of the seedlings noted.

Two soil temperatures 25° and 35° C. were used, and 15 kernels from each ear. The data are presented in table II.

In table II it is manifest that all ears which showed infection were not necessarily devitalized for many of them showed a perfect stand. Some ears showed the effect of the fungus by producing weakened seedlings, however, while others showed a high percentage of dead seed and still others had both kinds of injury represented. The effect of tem-



Fig. 6. Interior of kernel attacked by *Basisporium* showing disintegrated and blackened germ.

TABLE II. EFFECT OF *BASISPORIUM GALLARUM* AS SHOWN BY RESULTS ON 32 VISIBLY INFECTED EARS AT SOIL TEMPERATURES OF 25° TO 35° C.

Ear no.	25° C.		35° C.		Ear no.	25° C.		35° C.	
	Percent		Percent			Percent		Percent	
	Stand	Weak	Stand	Weak		Stand	Weak	Stand	Weak
1	60	6	47	40	9	33	6	53	6
2	93	..	87	..	10	83	..	92	13
3	100	..	87	..	11	93	..	100	27
4	87	..	100	..	12	87	..	100	46
5	80	..	87	6	13	87	..	87	46
6	100	..	100	..	14	80	20	67	6
7	93	..	93	53	15	60	..	93	40
8	80	13	67	6	16	60	..	80	27
17	100	6	80	13	25	87	..	80	..
18	87	..	100	..	26	93	..	93	..
19	93	..	100	..	27	73	40	87	40
20	100	13	100	..	28	87	..	100	6
21	60	..	87	..	29	80	6	93	33
22	100	..	93	..	30	87	13	87	..
23	100	13	100	..	31	80	6	73	13
24	80	46	93	20	32	87	6	87	..

perature will be discussed in detail later, but it may be pointed out here that the injury to germination was greater at the lower temperature. The total stand for the 25° C. plot was 83 percent as compared to a stand of 87 percent in the plot that was held at 35° C.

In order to test further the effect of *Basisporium gallarum* on vitality, seed from a similar lot of ears was planted in the field May 17, 1924. The percent stand and percentage of weak plants in this plot are given in table III.

It will be noted in this table that the different ears again present different degrees of injury as indicated by stand and weak plants. The average stand in the field was 63 percent for seed from infected ears against 82 percent stand in checks or healthy ears. This difference marks a distinct loss in stand which is reflected directly in the yield. Weakness and slowness of growth accompanies poor germination and in a number of instances a distinct blighting of the seedling occurred due to rotting off of the young plant near the seed.

The lower temperature of the field soil in May as compared to the greenhouse soil temperatures suggests that soil conditions at time of germination influence the degree of loss resulting from planting infected seed. Brief tests with infected seed from the same sample in warm and cold soil resulted as follows:

Infected seed in soil at 25° C. = 96 percent stand
 Infected seed in soil at 10° C. = 50 percent stand.

These data corroborate the findings at 25° and 35° C. in the greenhouse as reported in table I.

In the case of *Basisporium* dry rot, the decrease in stand is accompanied by the presence of weakness in the plants that do

TABLE III. PERCENT STAND IN FIELD RESULTING FROM BASISPORIUM INFECTED SEED.

Row no.	Basisporium dry rot ears		Healthy ears	
	Stand percent	Weak plants percent	Stand percent	Weak plants percent
1	80	23	77	6
2	84	11	86	8
3	75	20	86	7
4	66	11	87	7
5	70	19	87	13
6	70	4	81	6
7	80	10	74	10
8	60	14
9	51	7
10	52	8
11	77	20
12	62	22
13	86	11
14	73	14
15	46	13
16	60	19
17	46	6
18	81	8
19	29	3
20	61	8
21	65	19
22	68	17
23	35	16
24	48	14
25	43	7
26	65	8
27	91	10
28	52	20
29	30	10
30	80	16
31	66	8
32	54	20
33	77	16
34	30	7
35	61	11
36	75	11
37	78	11
38	46	7
39	75	10
40	71	13
41	74	13
42	88	8
43	74	7
44	90	11
45	75	13
46	18	4
47	77	14
48	42	7
49	74	13
50	36	8
Average	63	..	82	..

grow. It is evident in tables I and II that when a weakened condition exists, certain of the seed grow slowly and are out-distanced by others. Just what the history of these weak plants is, will be considered in future experiments.

MORPHOLOGY AND GERMINATION OF SPORES

The description of *Basisporium gallarum* by Molliard is quite complete and little can be added to it. However, the published figures of the fungus as found on corn are not very adequate in

identifying it as the same organism that Molliard described. The fungus is characterized by a sparse white mycelium bearing black sub-spherical spores on basidia-like sporophores. These spores, averaging 13 to 18 μ in diameter are almost spherical, but are slightly flattened at the point of attachment (fig. 7). They possess a thick black exospore which, when broken, allows the exit of a thin, light colored endospore. The pigmentation of these spores is intense black and the only agent so far found that even partially clears them is boiling nitric acid. On treatment with this reagent, the color is somewhat removed so that the cell contents and character of the spore wall may be seen. Along the edge of the spore what appears as a double line may be seen extending half way around the spore, constituting a suture which opens on germination, permitting emergence of the germ tube. Two germ tubes are usually sent out, not a single bifurcated tube as described by Molliard. Their shape and length depend upon the time, temperature and the medium in which they are germinated.

The development of the spore is shown in fig. 8. The spore usually arises from a basidia-like sporophore and at first it is hyaline or granular, becoming black on maturity.

The spores of *Basisporium gallarum* germinate with difficulty in water. Molliard and Arzberger report having observed germination, but they carried their investigations in this direction no farther. Ramsey noted that the spores germinated poorly in water, and found that tomato pulp gave much better results. He

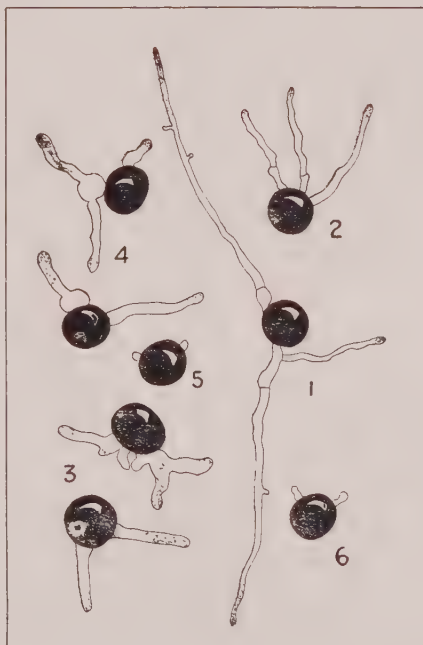


Fig. 7. Germinating spores of *Basisporium gallarum*. Normal germination in tomato juice five hours, 25-40° C. 2. Germination in H₂O—germ tubes alternated—25-40° C. 3. Germination in tomato juice after 2-3 hours 25-30° C. 4. Germination in tomato juice five hours, 35° C. 5. Germination in tomato juice just starting. 6. Germination in tomato juice five hours 15° C.

did not understand the true significance of this reaction and merely records the fact. The poor germination obtained in

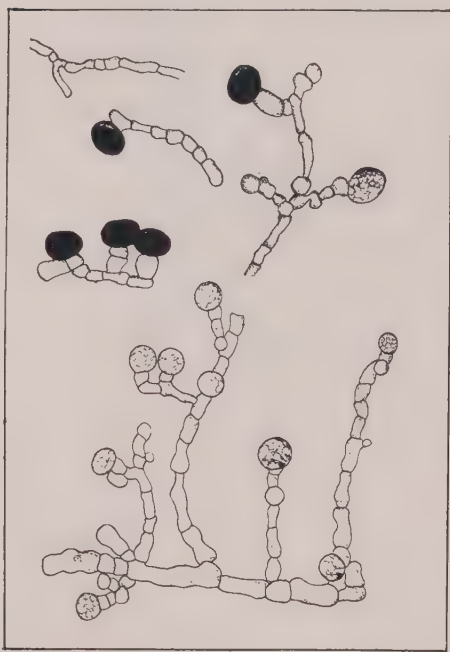


Fig. 8. Mycelium and spores of *Basisporium gallarum*. Vegetative hyphae and fertile hyphae showing character of sporophores and mature and immature spores.

water, coupled with Ramsey's results with tomato pulp led to experiments with tomato and other vegetable and fruit juices. In all of these, the germination was nearly perfect while in the checks in water, there was scarcely any germination.

Experiments with the spores of *Basisporium gallarum* in water cultures in the presence of tomato pulp revealed striking results. In repeated tests, wherever tomatoes were placed in the same chamber with the water drop cultures, 100 percent germination resulted. The checks in tap water, water distilled over block tin, and water

double distilled over glass showed no germination.

Other fruit and plant tissues as follows were also tried out: apple peel, apple pulp, orange peel, banana peel, verbena, nasturtium, lily, lilac flowers and leaves of geranium. In all instances, stimulation was obtained, equal or nearly equal to that with tomato.

Other workers have noted this condition with other fungi: notably Brown (2 and 3) working with *Botrytis cinerea*, and Leach (10) with *Colletotrichum lindemuthianum*. They found that the mere presence of host tissue in the same atmosphere as the drop culture influenced germination. Noble (12) found that distillates of plant tissues stimulated the germination of spores of *Urocystis tritici* and suggests that growing roots of the host plant where they are near spores may have a stimulating effect

on their germination. Griffiths (9) with the same fungus records the stimulating effect of germinating seeds on spore germination. As these tissues are odoriferous it was thought that some aromatic volatile substance might be instrumental in producing the stimulation of spore germination of *Basisporium gallarum*. Consequently several aromatic substances were used in minute amounts, namely: geraniol, citral, benzaldehyde, methyl alcohol, ethyl alcohol, ethyl acetate, acetic acid and ether. In repeated trials, the results were negative.

It is known that *Basisporium gallarum* germinates and grows in the cavity of the leaf sheath clasping the stalk in the presence of pollen grains. Similar conditions were duplicated in cultures of the spores. It was found that a few corn pollen grains germinating in the culture drop with the spores stimulated germination. Spores of other fungi had a like effect.

Later the effect of plant tissues in the same culture chamber, but not in the drop of water, was tried. Corn leaves, nasturtium leaves, Tradescantia leaves, cucumber leaves and pieces of corn stalk repeatedly produced strong germination under these conditions, while the checks in water drops in other closed vessels failed to germinate. The action of these tissues rather precludes the possibility of an effect from any specific volatile substance such as might emanate from some aromatic fruit.

Experiments endeavoring to explain this phenomenon on the ground of osmotic pressure relations gave negative results, as did trials on the differences in surface tension of the spore bearing drops. The hypothesis that the acid in tomato juice might be the active agent was suggested by the fact that the spores germinated as readily in orange and lemon as they did in tomato juice; results which parallel those obtained by Thiel and Weiss (15) in their use of citric acid in furthering germination of teleutospores of *Puccinia graminis*. The acids present in tomato as listed by Wehmer (16) are: citric 0.9 to 0.69 per cent; oxalic 0.001 percent; malic 0.48 percent; tartaric, succinic and salicylic acid, each a trace. The use of dilute solutions of these acids, as media for germinating spores, also gave negative results.

STIMULATING EFFECT OF CARBON DIOXIDE (CO₂)

The most universal volatile substance produced from plant tissue is CO₂. It is given off not only by leaves and flowers, but also by germinating seeds. Tashiro (14) has shown that this gas also is generated rapidly in large amounts by cut tissues. It is easy, therefore, to assume that in the germination experiments on spores of *Basisporium gallarum* where plant tissues were included in the same chamber with the drop cultures that the CO₂ generated by these tissues might be active in stimulating germination.

In order to demonstrate whether or not CO_2 from plant tissues is the active agent in the germination of the spores of *Basisporium gallarum*, water drop cultures were made with the following plant tissues in the chambers with the drops: orange peel, apple peel, verberna flowers, geranium leaf, nasturtium flowers, tobacco flowers, rose petals, apple pulp, crushed tomato fruit, whole tomato fruit, green corn leaf and cucumber leaf. In all these cultures germination was profuse, while the water check showed only a trace. A parallel set of cultures were run at the same time under the same conditions using the same plant tissues. In the bottom of this second set of chambers, however, a solution of barium hydroxide was placed, in an amount in excess of that necessary to take up the CO_2 given off by the plant tissue. In none of these cultures was germination better than in the checks. In all these culture chambers where barium hydroxide was used a heavy precipitate of the carbonate resulted after standing in the presence of plant tissues.

Further, where tomato pulp stood in a closed chamber for five or ten minutes and the air was then exhausted into a second closed chamber containing water drop cultures of *Basisporium gallarum*, germination was stimulated as in cases where the tissue remained in the same chamber with the drop. However, where the air from the tomato tissue was passed thru two washes of barium hydroxide before entering the culture chamber, no germination resulted, while checks in which the air was not passed thru barium hydroxide showed profuse germination.

Repeated tests, as above, indicate the agency of CO_2 in the stimulation of the germination of *Basisporium* spores by plant tissue. This is further borne out by the actual use of CO_2 . Carbon dioxide direct from a generator and also washed CO_2 were passed into chambers containing drop cultures. After standing the usual period used in other germination tests profuse germination resulted. The exact amount of CO_2 necessary for stimulation was not determined, tho it was found that an excess of CO_2 to the exclusion of oxygen prevented germination.

In this connection it might be noted that where spores are left in water drop cultures 48 to 64 hours, a fairly good percent of germination results. In the ordinary atmosphere there is approximately .03 percent CO_2 . It would appear then that if enough time is given, the culture drop can absorb CO_2 sufficient to influence germination. In this connection it is of interest to consider the reaction of dry spores. If dry spores are placed in a nearly saturated atmosphere they collect a film of moisture over the surface. This film can readily be seen with the microscope. Furthermore, spores covered with such a film germinate profusely in an atmosphere containing the normal amount of CO_2 . This suggests that where only a film of water surrounds

TABLE IV. INFLUENCE OF TEMPERATURE ON GERMINATION AND GERM TUBE GROWTH OF SPORES OF *BASISPORIUM GALLARUM*

Temperature.....	5° C.	10° C.	15° C.	20° C.	25° C.	30° C.	35° C.	40° C.
Percent germination..	0	0	18	67	68	71	47	0
Length of germ tube..	0	0	30	50	150	115	70	0

the spore, the amount of CO_2 as found in normal air is sufficient to stimulate germination. Possibly the greater surface exposed by the film facilitates the absorption of CO_2 and its transmission to the spore. Another possible explanation is, of course, that the CO_2 is converted into carbonic acid in the spore and changes the hydrogen ion concentration. The reverse of these experiments was also tried. Dry spores were placed on dry slides in a saturated atmosphere free of CO_2 . No germination resulted, while in control chambers where the CO_2 content was the same as in normal atmosphere, the germination percent was high. From the data at hand it is evident that CO_2 greatly stimulates the germination of spores of *Basisporium gallarum*; likewise, the accelerating effect of plant tissues on the germination of these same spores is due to the production of CO_2 by such tissues.

RELATION OF TEMPERATURE TO SPORE GERMINATION

In addition to these relations of CO_2 and plant tissue to germination, the temperature relations of any fungus are obviously of great importance, both from the standpoint of germination and of growth. In order to determine this relationship, drop cultures were made and held for five hours in electrically controlled chambers at temperatures ranging from 5° C to 40° C.

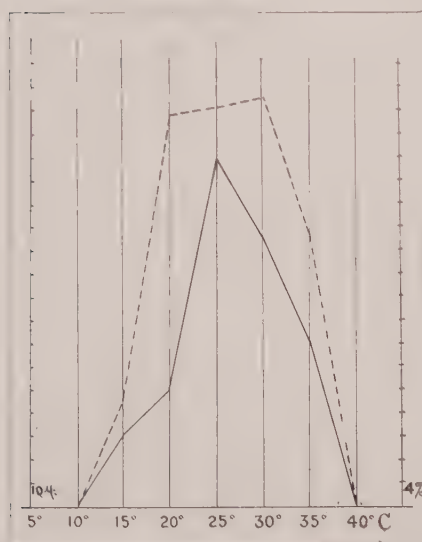


Fig. 9. Germination and growth of spores of *Basisporium gallarum* at different temperatures. Continuous line—growth of germ tube in microns. Broken line—percent germination.

That the spores might be readily examined, the drops were placed on the upper side of a clean glass slide and spores stirred into the drop. In this way the spores settled to the bottom of the shallow drop and were evenly distributed. If hanging

drops are used, the spores clump together to such an extent that the degree of germination cannot be distinguished. The slide

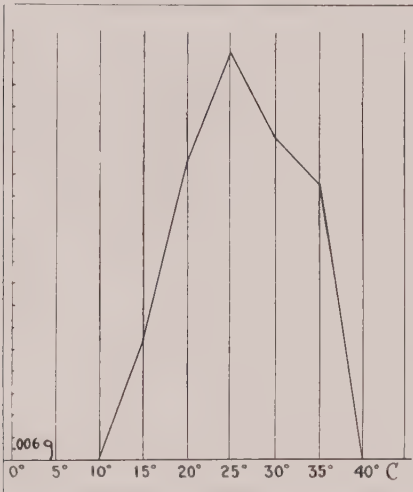


Fig. 10. Degree of mycelial growth of *Basisporium gallarum* at different temperatures expressed in grams dry weight.

with culture drop of green tomato juice on it was then placed on a moist filter paper in a petri dish and covered with a second slide having blocks of glass cemented to the ends, and the petri dish closed. Germination is slow, requiring three to five hours, the latter time being used in the experiment. The results are given in table IV.

In table IV, representing counts on 90 cultures, the relation of growth of germ tube to temperature is shown to be practically the same as that obtained in flask cultures. The optimum determined is in the neighborhood of 25° C. The maximum and minimum temperature at which growth takes place is 35° and 15° C., respectively. In the case of germination, however, little difference exists within a wide range of temperature and it would appear from the data at hand that temperatures within the ordinary ranges of growth have little influence on actual germination. Figure 9 shows that the germination curve becomes flattened between 20° and 30° C. This discrepancy suggests the entrance of a factor other than temperature, due perhaps to some content of the tomato juice.

RELATION OF TEMPERATURE TO MYCELIAL GROWTH

In addition to the temperature of germination, relation of temperature to the development of a fungus is usually of importance in understanding its nature and its relations to the host. *Basisporium gallarum* is no exception. In studying the effect of temperature on growth of *B. gallarum*, electrically controlled ovens were used and the fungus was grown on modified Pfeffer's solution containing M dextrose. Weighing of the fun-

gus felts developed was used as an indication of the growth obtained at the various temperatures.

TABLE V. DRY WEIGHT IN GRAMS OF FUNGUS FELTS AT DIFFERENT TEMPERATURES.

No. of cultures.....	5° C.	10° C.	15° C.	20° C.	25° C.	30° C.	35° C.	40° C.
45	0	0	.0363	.0894	.1215	.0837	.0750	0

Table V gives the average growth of the fungus in grams dry weight. The amounts represent averages of all cultures grown at the respective temperatures. Note that the optimum temperature for mycelial growth is 25° C. tho good growth occurs from 20° to 30° C. Forty degrees inhibits growth. This was also true of 10° C. or below; however, these temperatures do not kill the fungus because upon removal to the optimum temperature, growth is begun. The graph in fig. 10 further illustrates this temperature relation.

Supplementing the flask cultures, petri dish cultures were held at the same temperatures with similar results.

5° C.—no growth.

10° C.—no growth except a few short hyphae around original planting.

15° C.—sparse white growth—colony 1" diameter.

20° C.—white growth over entire plate—beginning to sporulate.

25° C.—culture spread over dish fruiting—black with spores.

30° C.—colony spread over dish, fruiting.

35° C.—slight growth 2" diameter—slight fruiting.

40° C.—no growth.

These results indicate the effect of temperature both on mycelial growth and upon spore production. Contrary to Arzberger's statement, the fungus fruits at the higher temperatures, tho as stated by Ramsey, sporulation is lessened at 35° C.

NUTRITIONAL REACTIONS

The action of *Basisporium gallarum* on specific tissues of the corn plant as noted in examining several thousand ears of corn and reported under symptoms suggested a study of the nutritional relations of the fungus.

TABLE VI. GROWTH OF *BASISPORIUM GALLARUM* ON MODIFIED .. PFEFFER'S MEDIA.

Nutrient used $\frac{M}{10}$	Average weight of fungus felts	Character of growth	
Starch	.0169 gm.	white	sparse
Dextrin	.0191	white	sparse
Cellulose	.0238	white	sparse
Glycerine	.1089	grey	thick
Alcohol	.0132	white	sparse
Dextrose	.0232	black	fruiting profusely
Maltose	.0439	black	" "
Levulose	.0312	dark at edges	
Sucrose	.0438	dark at edges	
Lactose	.0502	white	
Galactose	.0423	dark at edges	
Arabinose	.0230	white	sparse
Mannit	.0257	white	sparse

TABLE VII. GROWTH OF *BASISPORIUM GALLARUM* ON DIFFERENT MEDIA

Medium	Size of colony in inches	Reaction
Casein	2.	sparse growth—complete digestion
Amygdalin agar	0.	no growth
Cellulose agar	0	sparse growth—no visible digestion
Egg albumen agar	2.	digestion—sporulation
Peptone agar	2.5	vigorous growth
Litmus cream agar	.7	no acid reaction—profuse sporulation
Skimmed milk agar	2.	sparse growth—digestion and sporulation
Starch agar	1.7	slight digestion, halo
Asparagin agar	...	no reaction—sparse growth
Inulin agar	...	very slight growth—hyphae black

Both Arzberger and Ramsey record the ready growth of the fungus on a number of cultural media; however, in order to obtain a better idea of the food requirements than that furnished by the usual stock media the organism was grown on a modified Pfeffer's solution containing various nutrients as given in table VI.

In addition to these media the fungus was grown at its optimum temperature on a series of nutrients as described by Crabill and Reed (5). These media were poured in plates and the size of colony growth and reaction of the fungus noted.

Table VII shows that *Basisporium gallarum* is capable of using a wide range of nutrients and is able to secrete several enzymes. Good growth is obtained on casein, peptone and albumen. Starch dextrin and the various sugars are also used, while glycerine produces a profuse vegetative growth. The positive reaction of the fungus on the more nitrogenous food materials is of interest considering its effect on the corn kernel. The fungus destroys the embryo of the kernel rather than the starch endosperm. Illustration of this is given in fig. 6.

Cellulose is acted upon, but not as vigorously as might be expected of an organism that is reputed to grow in the cob of corn. Its growth on proteins and fats is a great deal more vigorous, as is also its growth on dextrose and maltose. In fact, these are the only materials on which it was found to sporulate. Its action on cream agar is curious. This medium is filled with tiny drops of butterfat in suspension; these butterfat drops were surrounded by hyphae which became black and resembled pycnidia. In this respect, they were very deceptive.

In sterilized field soil the fungus makes only a slight growth. In the many cultures made, only a few showed visible growth, a fine web of the mycelium of the fungus being apparent in the interstices of the soil.

INFECTION STUDIES

There are two periods in the life history of the corn plant when it is most susceptible to attack by *Basisporium gallarum*; first, in the seedling stage, sprouting, and secondly, at the time

of maturity. Infection is local on the corn plant at the time of maturity, accompanying excessive moisture. In 276 inoculations made on roots, nodes, internodes, leaves and ears, Arzberger was unable to obtain infection. Neither did he obtain infection on seedlings. On moist mature ears, however, he obtained growth of the fungus tho on green growing ears no growth resulted from his inoculations. Ramsey readily obtained infection on tomato when the skin of the fruit was injured. In this respect *Basisporium gallarum* resembles certain of the fungi causing apple and other fruit rots.

In field experiments with *Basisporium gallarum* on corn, it was found that the fungus does not migrate in the stalk or seldom enters the corn plant from the soil by way of the roots. Plantings from 1,200 stalks node and internode from the ground up to the ear, disprove any such assumption.

Infection experiments in the field on seedlings gave negative results. Healthy tested seed was planted in the field at different dates and germinated under varying weather conditions. The seed was planted by hand, each seed in contact with a mass of *Basisporium* inoculum, previously grown by culturing the fungus in mass on sterilized oats. Table VIII records the results.

In table VIII the results of planting healthy seed in contact with the fungus show no effect. Tho the four plantings were made at about one week intervals and experienced different growing conditions, only the one planted May 21 showed any decrease in stand from that of the check plot and this decrease was not significant.

One hundred seven of these plants were dug when a foot tall and the roots washed. The roots were entangled in the mass of original inoculum, but with the exception of a few lesions on some of the smallest roots, no effect could be seen. The remaining plants grew to maturity without any evidence of root injury.

RELATION OF WEAKNESS TO BASISPORIUM INFECTION.

In field plots in six counties, observations were made to determine whether a weakened condition of a corn plant rendered it especially susceptible to *Basisporium* dry rot.

TABLE VIII. EFFECT OF *BASISPORIUM GALLARUM* ON SEEDLINGS FROM CLEAN SEEDS.

Date planted	Number of plants	Percent stand
May 15	93	95
Check	270	94
May 21	117	88
Check	102	96
June 1	51	96
June 8	51	100
Check	117	96

TABLE IX. THE RELATION OF WEAK PLANTS TO INFECTION BY *BASISPORIUM GALLARUM*.

Location of plot	Number of 1/100 acre rows	Number of replications	Average yield per acre bu.		Number of rotted ears	
			Healthy	Weak	Healthy	Weak
Dallas county	48	3 series	59	55	58	69
Grundy county	42	3 "	66	52	60	62
Hardin county	66	3 "	58	49	208	214
Henry county	54	3 "	55	48	75	86
Marion No. 1	72	3 "	66	44	76	50
Marion No. 2	48	2 "	57	45	46	46
Webster	30	3 "	47	35	87	82
Average			58	47	90	87

The corn in the plots in the various counties was from seed part strong and part diseased with *Diplodia* dry rot. The diseased and healthy corn was planted in 1/100 acre adjacent plots. The plants from the diseased seed were weak. The results of *Basisporium* infection on these weak and healthy plants is shown in table IX. From the averages recorded (table IX) there is some evidence that the condition of vigor of the corn plant plays a part in *Basisporium* infection. Reduced to an equal yield basis, the results indicate 16 percent more *Basisporium* infection on ears of weak plants than on vigorous plants. Similar evidence was obtained on corn, Iodent variety, grown on the Iowa experiment station farm.

TABLE X. INFECTION OF DIFFERENT VARIETIES WITH *BASISPORIUM* DRY ROT.

Variety		Source	Percent rot	Maturity
1	Yellow dent	Grundy Center, Ia.	18	
2	Reid's Yellow Dent	Mt. Pleasant, Ia.	16	
3	White?	Knoxville, Ia.	15	
4	Wimple's Hybrid	Canada	28	Early
5	Iowa Ideal	Indianola, Ia.	22	
6	Silver King	Ackley, Ia.	20	
7	Boone Co. White	Lacy, Ia.	22	
8	White?	Indianola, Ia.	2	
9	Calico	Harcourt, Ia.	34	Early
10	Reid's Yellow Dent	Adel, Ia.	2	Late
11	Silver King	Osage, Ia.	16	
12	Wimple's Yellow Dent	South Dakota	24	
13	White?	Hamilton, Ia.	20	
14	Reid's Yellow Dent	Pleasantville, Ia.	15	
15	Murdock	Peters, Minn.	35	Early
16	Murdock	Mitchell, Ia.	33	Early
17	Silver King	Ackley, Ia.	19	Early
18	N. W. Dent	St. Paul, Minn.	13	Early
19	Lexal Tender	Elliott, Ia.	36	Early
20	Silver Mine	Elliott, Ia.	32	Early
21	Improved Reid's	Elliott, Ia.	0	Late
22	Silvermine x Silverking		12	
23	Johnson Co. White	Ottumwa, Ia.	3	
24	Yellow dent	Knoxville, Ia.	20	
25	Yellow dent	Faulkland, Ia.	25	Early
26	Boone Co. White	Univ. of Missouri	0	Late
27	Boone Co. White	Univ. of Missouri	0	Late
28	Gold Mine	Iowa Seed Co.	16	
29	Gold Mine	Iowa Seed Co.	20	
30	White		44	Early
31	White		9	
32	White		25	
33	Ohio White	Mt. Pleasant, Ia.	0	Very late

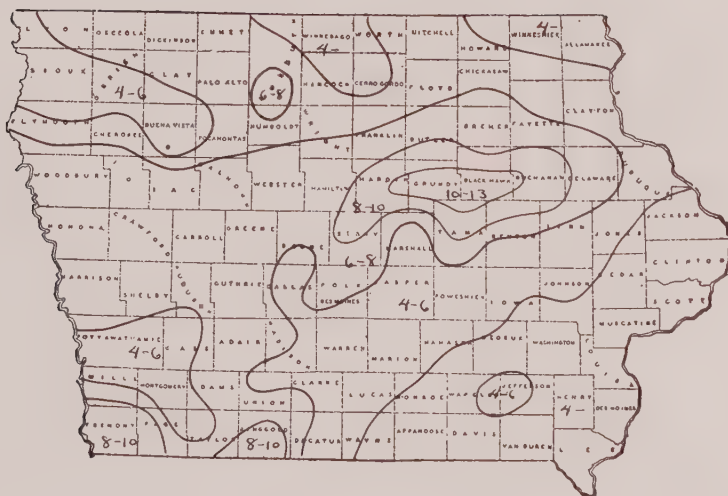


Fig. 11. Rainfall map of Iowa, August, 1923.

Again it may be that this reaction is due largely to the time of maturing of the weak plants as compared to the healthy ones.

In the county plots, corn from a large number of sources was planted. Varying degrees of *Basisporium* dry rot were found on the different strains; also the times of maturity of the strains varied. On the more uniform ripening plot of Iodent on the experiment station farm, the amount of *Basisporium* infection was quite uniform for the different rows.

RELATION OF TIME OF MATURITY ON BASISPORIUM INFECTION

The relation of time of maturity to infection with *Basisporium gallarum* is most striking where widely different varieties are considered. In table X are given the percentages of *Basisporium* dry rot as found on harvested ears from 33 strains of corn, planted in 25 hill rows on the Iowa experiment station farm in 1923. This planting was in a continuous plot surrounded by other corn.

These data suggest that time of maturity influences infection. Under the conditions prevailing in 1923, the year this corn was grown, the heavy precipitation in August and the first part of September was conducive to *Basisporium* infection on the early maturing varieties.

Plants that matured late did not offer such opportunity to the fungus and reached their most susceptible condition at a time of little rainfall. It is obvious that conditions from year to year

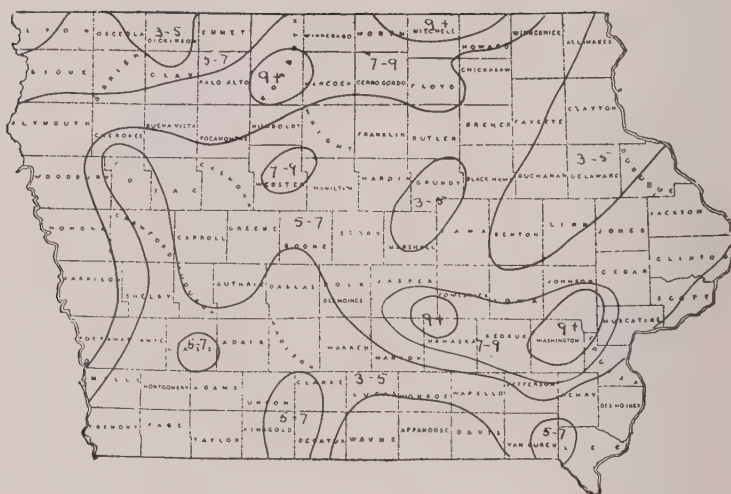


Fig. 12. Rainfall map of Iowa, September, 1923.

may vary so that the combination of maturity and precipitation may not coincide with the result that little or no *Basisporium* dry rot develops.

RELATION OF WEATHER TO INFECTION.

A comparison of weather conditions for 1922 and 1923 showed the most rainfall during August and September of 1923. It was during this period that *Basisporium* dry rot developed. No such general and heavy rainfall occurred in 1922, altho local areas experienced excessive amounts of moisture.

In Iowa as a whole, the precipitation for August, 1923, (fig. 11) was 5.42 inches, the heaviest rainfall for that month since 1903, exceeding the normal by 1.74 inches. September (fig. 12) with 5.79 inches, was 2.43 inches above normal. The rainfall for these two months combined was 11.21 inches, which was unusually heavy. *Basisporium* dry rot was severe while in other years when the precipitation was low during this period the amount of disease was practically nil.

Incidentally, it is interesting to note that in 1911, a year when *Basisporium* dry rot was very prevalent in Ohio (1), the weather reports of that state for August and September show excessive rainfall in both months.

These facts led to the conclusion that moisture is the chief factor in the development of the disease, a conclusion which is further confirmed when a comparison is made between the prevalence of the *Basisporium* dry rot over the state and the amount

TABLE XI. PERCENT OF BASISPORIUM DRY ROT AT VARIOUS STATIONS COMPARED WITH RAINFALL.

County	Average percent Basisporium	Rainfall in inches	
		August	September
Dallas	5.4	4-6	3-5
Grundy	9.4	10-13	3-5
Hardin	16.8	8-10	5-7
Henry	7.9	4-	3-9
Marion	4.0	4-6	3-9
Webster	16.2	6-9	7-9

of rainfall in various localities. For instance, in the southern part of Marshall county and in Jasper county the rainfall during the critical period for the disease was very light compared to that in the rest of Iowa. Only a slight amount of *Basisporium* dry rot was found in the area. For miles the fields visited showed no trace of the disease, whereas, in the wetter areas of the state the disease was much more prevalent as stated above. More definite data were gathered in a series of county test plots. Corn was obtained from a number of farmers in each county and seeded in plots of 1/100 acre rows in three replications. Table XI gives a direct comparison of the percentage of *Basisporium* dry rot present and the rainfall.

The correlation with the precipitation for August and September is evident, the greater amount of rot being coincident with the greater rainfall. The discrepancy occurring in the figures for Grundy county, which had a rainfall equally as heavy as Hardin, but had a lower percent of *Basisporium* dry rot, was due to the later maturing of the Grundy county plot.

SPREAD OF *BASISPORIUM GALLARUM* IN CRIB

The prevalence of moldy corn in cribs in certain seasons raises the question, does *Basisporium gallarum* spread in the crib? During seasons of extreme moisture, as in the fall of 1923, little opportunity was offered for the corn in the center of cribs to dry properly. A number of such instances were observed; in all cases, however, the molding was due to species of *Penicillium*. Tho *Basisporium gallarum* was also found, no evidence of spread of that fungus was observed. In experimenting with the spread of the mold in moist chambers, it was found that ears slightly affected with *B. gallarum* or sound ears on which very small pieces of the fungus were placed, became infected with the fungus if kept in a saturated atmosphere. No infection resulted, however, from inoculum introduced into a mass of damp ears. A bushel of sound, moist ears was placed in a closed iron can and a liter of *Basisporium* inoculum (soaked sterile oats used as media) placed in the center in contact with the ears. After two months the cap was opened, but little evidence of spread of the fungus was found, tho the ears were moist, and *Aspergillus niger* and species of *Penicillium* grew on many of them.

OVERWINTERING.

The nature of the thick-walled spores of *Basisporium gallarum* suggest their resistance to weathering and longevity in the soil. In germination tests of spores gathered on corn stalks in the field, it was found that spore from the previous season readily germinated with vigor and in as high percentage as those carried over winter under shelter. Whether they will live in the soil more than one year, as is the case with *Diplodia zeae*, is not known.

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